Insulin receptor substrate-2 – a new candidate gene for NIDDM?

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The pathogenesis of non-insulin-dependent diabetes mellitus (NIDDM) includes the development of insulin resistance and, at a later stage, impairment of β-cell response. In the past few years, different elements, including the proto-oncogene, Shc, and the insulin receptor substrate (IRS) proteins, have been found to have important roles in the insulin signaling network. Whithers et al. (1) have now demonstrated that IRS-2 is crucial for intact insulin and β-cell function. By knocking out the IRS-2 gene, they created a mouse model showing the typical features of human NIDDM, including insulin resistance and β-cell dysfunction. Understanding of this discovery is aided by scrutiny of the current model of insulin signaling and the outcome in the IRS-1 knockout mouse (reviewed in (2) and (3)).

Insulin binding to its αβ2-heterotetrameric receptor leads to autophosphorylation and activation of the tyrosine kinase of the β-subunit. Even though the subsequent phosphorylation of and interaction with intracellular proteins activates a complex signaling network, two major pathways can be distinguished that diverge at the level of the IRS and Shc.

Phosphorylation of Shc by the insulin receptor tyrosine kinase leads to stimulation of the mitogen-activated protein kinase (MAPK) pathway, which is involved in mitogenesis, but also in glucose clearance, and ultimately results in glucose transporter 1 (GLUT1) translocation to the cell membrane and glucose synthesis via stimulation of the ribosomal S6 kinase pp90rsk. The link between Shc and MAPK is provided by the link between Shc with IRS, such as PI3-K or the proteins fyn, syp, nck and GLUT4 which, in contrast to the ubiquitous GLUT1, is involved in mitogensis, but also in glucose clearance, and ultimately results in glucose transporter 1 (GLUT1) translocation to the cell membrane and glycogen synthesis via stimulation of the ribosomal S6 kinase pp90rsk. The link between Shc and MAPK is provided by the autophosphorylation and activation of the MAPK kinase-kinase (MEK) and finally the MAPK.

The other major pathway consists of tyrosine phosphorylation of members of the IRS family, leading to activation of phosphatidylinositol 3-kinase (PI3-K). This enzyme plays an important part in a variety of biological events including mitogenesis, vesicle sorting, receptor internalization and glucose homeostasis. Stimulation of PI3-K results in protein synthesis via activation of the pp70s6k, and in translocation of GLUT4 which, in contrast to the ubiquitous GLUT1, is exclusively responsible for glucose uptake into insulin-sensitive tissues. Most elements that interact directly with IRS, such as PI3-K or the proteins fyn, syp, nck and Grb-2/Sos, contain so-called SH2 (Src homology-2) domains that bind to the tyrosine phosphorylation sites of the IRS protein. As mentioned above, Grb-2 and Sos are also involved in linking Shc with p21ras, and thereby confer crosstalk of the two pathways by activating ras in an IRS-dependent but also an independent fashion. However, p21ras also seems to activate PI3-K directly, providing an additional interaction with IRS signaling. There are also elements lacking SH2 domains (SV40 large T antigen, 14-3-3 proteins, β-integrins) that bind IRS independently of phosphotyrosine but the role of these elements, and those of fyn, syp and nck, remain unknown.

Signal specificity in the Shc or IRS system seems to be conferred by distinct tissue distribution or phosphorylation patterns of the signaling proteins as, otherwise, redundancy is remarkable. In addition to the insulin receptor, a variety of other cell membrane receptors (insulin-like growth factor 1, growth hormone, interleukine (IL) and interferon receptors) use the same intracellular substrates, and several Shc and IRS isoforms have been identified.

IRS-1 was first described as a portion of a 185-kDa phosphoprotein (pp185) isolated from insulin-stimulated hepatoma cells by means of precipitation by anti-phosphotyrosine antibodies. The surprising facts that the IRS-1 knockout mouse (IRS-1−/−) was not diabetic and had only mild insulin resistance and mainly a 50% growth retardation, suggested the presence of compensatory elements. In fact, pp185 contains a high-molecular-weight portion first noted after IL-4 stimulation and therefore designated 4PS (IL-4 phosphoprotein substrate), which was then cloned from myeloid cells and, because of homology with IRS-1, renamed IRS-2 (4). In common with IRS-1, IRS-2 stimulates PI3-K and provides binding sites for IRS signaling. The IRS-2 knockout mice (IRS-2−/−) had a phenotype very distinct from that of IRS-1−/− mice. Accordingly, IRS-2 expression seems to be upregulated in these IRS-1 mutant mice (5).

In order to define the role of IRS-2, Whithers et al. (1) inactivated the IRS-2 gene by homologous recombination. The IRS-2 knockout mice (IRS-2−/−) survived, and lack of IRS-2, in addition to normal IRS-1 expression, was confirmed in the tissues of these mutant mice. Interestingly, IRS-2−/− had a phenotype very distinct from that of IRS-1−/− mice. Compared with those in wild-type mice, plasma glucose concentrations were already increased 3 days after birth: at 6 weeks of age the mice showed glucose intolerance and fasting hyperglycemia, and at
10 weeks they were diabetic with polydypsia and polyuria, and died of hyperosmolar coma with no ketoacidosis. Using insulin tolerance tests and the euglycemic hyperinsulinemic clamp and tracer technique, Whithers et al. were able to demonstrate that the animals suffered from severe insulin resistance affecting the liver and skeletal muscle. While expression of the insulin receptor and its phosphorylation remained normal in IRS-2−/−, the insulin-stimulated increase in PI3-K activity was markedly reduced, providing a possible explanation for the disturbed glucose homeostasis.

In addition to insulin resistance, the IRS-2 knockout mice were β-cell deficient compared with wild-type mice, and even more so compared with IRS-1−/−, which demonstrate a clear compensatory increase in β-cell mass. Interestingly, IRS-2 was found to be co-localized with pancreatic β-cells and ductal islet precursor cells, suggesting that its absence in the IRS-2−/− mice might cause a defect in the proliferative response of the islet to hyperglycemia. These findings are in contrast to those with other mouse models of insulin resistance, but in agreement with the findings in human NIDDM, and may explain the progression to overt diabetes in the IRS-2−/− mouse (1, 5).

In human NIDDM, IRS-2 expression seems to be normal (6); however, this does not preclude any mutations or functional impairment, or any defects of IRS-2-specific downstream elements.

The distinct phenotypes of IRS-1−/− and IRS-2−/−, together with in vitro studies, suggest that IRS-1 might be more involved in the mitogenic effects of insulin and in protection from apoptosis affecting proliferation and growth, whereas IRS-2 has more metabolic functions. Obviously, IRS-2 seems to have a more critical role in insulin signaling, but it is not clear whether there is any interaction between IRS-1 and IRS-2 and one can speculate as to what the phenotype of a double knockout mouse (IRS-1−/−, IRS-2−/−) would be, if viable at all. Recently, third and fourth IRS isoforms (IRS-3, IRS-4) were cloned (7–9); their roles have yet to be established.

IRS-2, which seems to be crucial for β-cell proliferation, is engaged not only by the insulin receptor, but also by a variety of cytokine receptors. It is therefore tempting to speculate that an inadequate immune response competing for IRS-2 or impairing its function could cause a defect of β-cell proliferation or compensation during development or adult life and would thereby contribute to the susceptibility to diabetes.

Even though IRS-2 is a good candidate gene for NIDDM, there is still a long way to go before the mechanism of insulin resistance and the pathogenesis of NIDDM will be fully understood, and there may be more surprises before the causative elements for this common human disease are identified.

References
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